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Artificial Neural Network TSR for Optimization of Actinomycin Production

Marzieh Lamtar-Gholipoor[1,*], Soheil Fakheri[2], Mahmoud Alimoradi[2,3]

[1] Department of Chemical Engineering, Quchan Branch, Islamic Azad University, Quchan, Iran; marzieh.gholipoor@gmail.com.
[2] Department of Computer Engineering and Information Technology, Lahijan Branch, Islamic Azad University, Lahijan, Iran; fakherisoheil@iau.ac.ir; mahmoud.alimoradi@shafagh.ac.ir.
[3] Department of Computer Engineering, Shafagh Institute of Higher Education, Tonekabon, Iran; mahmoud.alimoradi@shafagh.ac.ir.

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Abstract

The optimization of industrial fermentation processes, particularly for the production of bioactive compounds like Actinomycin V, is essential for maximizing yield and cost-efficiency. This study introduces a hybrid approach integrating Artificial Neural Networks (ANNs) with the Trees Social Relations Optimization Algorithm (TSR) to optimize medium composition for the production of Actinomycin V by Streptomyces triostinicus. Traditional optimization techniques, such as Response Surface Methodology (RSM), often fall short of capturing the complex, non-linear interactions between medium components. By contrast, the ANN-TSR hybrid approach leverages the predictive power of neural networks and the robust optimization capabilities of TSR, inspired by the resource-sharing behaviors of tree communities. The study employed a Central Composite Design (CCD) to systematically vary concentrations of key medium components, with experimental data used to train the ANN. The TSR algorithm then iteratively refined the ANN model to identify optimal conditions, significantly increasing Actinomycin V yield from an initial 110 mg/L to 443 mg/L. This fourfold enhancement underscores the potential of combining advanced machine learning techniques with nature-inspired optimization algorithms to optimize complex bioprocesses. The methodology presented here offers a generalizable framework applicable to various industrial bioprocesses.

Keywords: Trees social relationship algorithm, Fermentation, Neural network modeling, Metaheuristic algorithm, Machine learning, Optimization.

1 | Introduction

Actinomycin V, a potent anti-cancer agent, effectively inhibits the colony formation of F5-5 Friend leukemia cells when produced via submerged fermentation. Optimizing the medium composition is crucial for enhancing the efficiency and cost-effectiveness of industrial fermentation processes. Experimental designs

Corresponding Author: marzieh.gholipoor@gmail.com
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must ensure uniform distribution of each component across the sample space to evaluate the influence of different medium components on antibiotic production. While traditional methods like the One-Factor-At-A-Time (OFAT) approach are commonly used, more sophisticated statistical and mathematical techniques, such as full and partial factorial designs, offer greater accuracy. The Central Composite Design (CCD) is often chosen for its orthogonality and rotatability, making it ideal for experiments aimed at developing mathematical models that link medium components with antibiotic yields [1], [2].

Response Surface Methodology (RSM) is typically employed to build these mathematical models. However, RSM's reliance on quadratic polynomials may not always capture the relationships' complexity. Machine learning techniques like Artificial Neural Networks (ANNs) can provide a more precise alternative. ANNs consist of layers—input, hidden, and output—with the input layer receiving normalized data, the hidden layer processing it through weighted connections, and the output layer generating the final predictions. The performance of an ANN is influenced by both the weights and the transfer function used within the network [3], [4]. *Fig. 1* shows the performance of ANN during the training process.

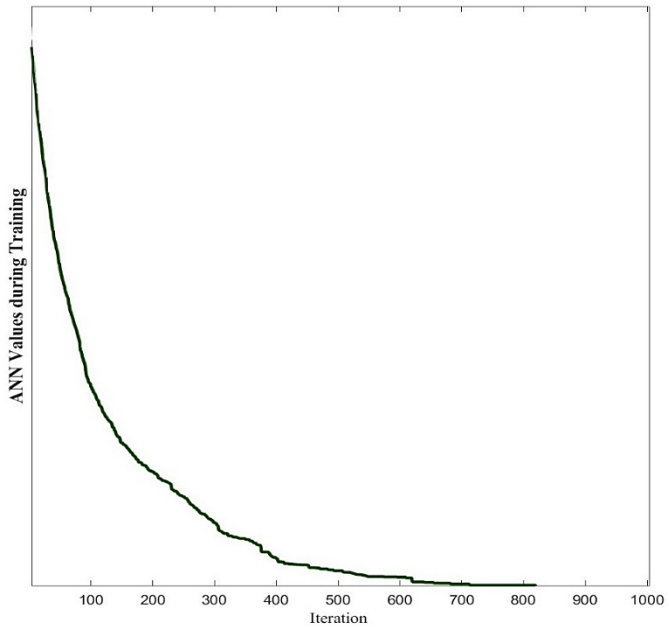

Fig. 1. Performance of ANN during the training process.

The Trees Social Relations (TSR) optimization algorithm can be used alongside ANNs to optimize medium composition. TSR, inspired by the natural interactions within tree communities, iteratively refines solutions by simulating trees' social relationships and resource-sharing behaviors. Through this process, TSR identifies the optimal concentrations of medium components to maximize antibiotic production. While the combination of ANNs and TSR has successfully addressed various optimization problems, it is not without limitations. The accuracy of these models heavily depends on the quality of the input data, and significant computational resources are required to prevent overfitting. Additionally, these methods may not always fully capture the effects of different medium components [5]–[7].

Despite these challenges, neural networks have proven effective in a wide range of applications, from autonomous flight control to medium design and optimization. In our study, we applied the ANN-TSR combination to optimize the production of Actinomycin V from Streptomyces triostinicus, a strain we isolated. Previous research has documented the production of Actinomycin V from various Streptomyces species, with a new isolate, Streptomyces MITKK-103, identified as a particularly potent producer. Our goal was to enhance the yield of Actinomycin V from our isolated strain by optimizing the medium composition. All neural network models and TSR optimization algorithms were implemented using MATLAB V.20, leveraging its advanced capabilities in numerical analysis, matrix computation, signal processing, and graphics [8], [9].

Table 1 presents the CCD analysis, showcasing the effect of five factors: MgSO4, NaCl, Glucose, Soybean, and CaCO3 on antibiotic yield, measured in mg/l. It includes observed antibiotic yields alongside predictions from ANNs and RSM for comparison. The table consists of 30 experimental runs, with varying concentrations of the five factors, demonstrating how different combinations influence the antibiotic yield while also comparing the accuracy of the ANN and RSM predictive models against observed results.

Table 1. CCD analysis of five factors, concentration units, antibiotic yield.

Runs	MgSO4 (g l−1)	NaCl (g l−1)	Glucose (g l−1)	Soybean (g l−1)	CaCO3 (g l−1)	Antibiotic Yield (mg l−1)	Observed	ANN Prediction	RSM Prediction
1	0.3	2.0	10	6.82	4.0	190	190	190.001	193.343
2	0.3	2.0	10	22.5	2.0	200	208.333	208.333	191.401
3	0.3	2.0	30	6.82	2.0	290	289.9	289.9	286.925
4	0.3	2.0	30	22.5	4.0	220	208.333	208.333	204.501
5	0.3	4.0	10	6.82	2.0	240	239.996	239.996	235.146
6	0.3	4.0	10	22.5	4.0	190	189.998	189.998	195.607
7	0.3	4.0	30	6.82	4.0	220	219.949	219.949	214.466
8	0.3	4.0	30	22.5	2.0	190	189.997	189.997	172.312
9	0.7	2.0	10	6.82	2.0	210	209.988	209.988	224.315
10	0.7	2.0	10	22.5	4.0	220	220.005	220.005	224.774
11	0.7	2.0	30	6.82	4.0	240	239.999	239.999	224.312
12	0.7	2.0	30	22.5	2.0	205	208.333	208.333	200.076
13	0.7	4.0	10	6.82	4.0	235	235.002	235.001	230.721
14	0.7	4.0	10	22.5	2.0	230	230.064	230.063	222.432
15	0.7	4.0	30	6.82	2.0	260	260	260	252.243
16	0.7	4.0	30	22.5	4.0	190	189.997	189.997	182.312
17	0.1	3.0	20	15	3.0	230	230.001	230.001	234.772
18	0.9	3.0	20	15	3.0	240	239.989	239.989	232.321
19	0.5	1.0	20	15	3.0	270	270.011	270.011	260.034
20	0.5	5.0	20	15	3.0	240	240.054	240.054	250.535
21	0.5	3.0	0.02	15	3.0	190	190.003	190.003	183.401
22	0.5	3.0	40	15	3.0	200	199.993	199.993	207.801
23	0.5	3.0	20	0.02	3.0	220	219.99	219.99	233.632
24	0.5	3.0	20	30	3.0	190	189.998	189.998	183.212
25	0.5	3.0	20	15	0.1	230	229.999	229.999	235.401
26	0.5	3.0	20	15	5.0	210	210	2100.001	203.012
27	0.5	3.0	20	15	3.0	255	255.654	255.653	235.789
28	0.5	3.0	20	15	3.0	240	239.521	239.521	235.789
29	0.5	3.0	20	15	3.0	240	239.521	239.520	235.789
30	0.5	3.0	20	15	3.0	230	239.521	239.520	235.789

2|Fermentation Process and Medium Optimization

A microorganism capable of producing Actinomycin-V, referred to as M4, was isolated from a pre-treated soil sample collected from an agricultural field. This strain was identified as Streptomyces triostinicus through 16S rRNA homology. It has been deposited in the Microbial Type Culture Collection (MTCC) in Chandigarh, India, under the accession number MTCC 8123. The strain's sequence is also available in the National Center for Biotechnology Information (NCBI) database with the accession number EU635725 [2], [10].

Submerged batch fermentations were carried out over six days at 180 rpm and 28°C in 250 ml Erlenmeyer flasks containing 50 ml of production medium. The medium composition included:

I. MgSO4·7H2O: 0.5 g/L.
II. (NH4)2HPO4: 0.5 g/L.
III. NaCl: 3 g/L.
IV. K2HPO4: 1 g/L.
V. Glucose: 15 g/L.
VI. Soybean meal: 10 g/L.
VII. CaCO3: 3 g/L.

The production medium was inoculated with 1% (v/v) of a 48-hour-old seed culture. The seed culture's medium composition and growth conditions were identical to those used for production, except for the 48-hour harvesting time. The concentrations of MgSO4, NaCl, glucose, soybean meal, and CaCO3 were adjusted based on the CCD to optimize the medium. The other medium components remained constant, with concentration ranges determined using results from the Plackett–Burman design and response surface experiments [2], [11].

Post-fermentation, the broth was centrifuged at 11,086 g for 20 minutes to separate the cells. The supernatant was then extracted with ethyl acetate and concentrated under vacuum. The concentration of Actinomycin V was measured by its UV absorption in methanol using a Perkin Elmer Lambda-25 UV spectrophotometer at 443 nm, specific to the phenoxazone nucleus. High-Performance Liquid Chromatography (HPLC) analysis was conducted using an analytical reverse-phase C-18 silica column (Lachrom) at a flow rate of 0.5 ml/min with a water–acetonitrile gradient on a Merck system [12].

A feed-forward back-propagation neural network was trained to optimize the media for antibiotic production using data from five variables specified in the CCD. The concentrations of the medium components were normalized using a log-sigmoidal function

$$\mathrm{m} = \frac{1}{1 + \exp(-\mathrm{n})}, \quad \textbf{(1)}$$

where (n) represents the medium component concentrations from *Table 1*, and (m) is the output used as the input for the neural network. The network was trained with data from twenty randomly selected experiments, while the remaining data were used for validation. The training performance, from the initial stages to the final adaptation, is shown in *Fig. 1*.

3 | Structure and Training of the Neural Network

The applied ANN model consisted of three layers of neurons:

I. Input layer: five neurons corresponding to the predictor variables.
II. Hidden layer: an optimized number of neurons.
III. Output layer: a single neuron.

The neurons are connected hierarchically, with the output from one layer serving as the input for the next. The learning process followed the back-propagation rule, where the error between the predicted and actual outputs was calculated and propagated backward through the network. This rule adjusted the weights in each layer to minimize the error, continuing iteratively until the error met a predefined criterion. One key advantage of using ANNs is that there is no need to know the exact form of the analytical function for the model. Unlike RSM, which requires fitting the model to a specific equation, ANNs do not require specifying the function type or the number of parameters [8], [13], [14].

4|Optimization with TSR Algorithm

The neural network optimization was carried out using MATLAB's 'treesocialopt' function. The input parameters for the TSR optimization algorithm TSR included:

I. Organism type: double vector.
II. Initial growth range: 1e−009—2.
III. Interaction fraction: 1.
IV. Elite count: 2.
V. Population size: 250.
VI. Resource sharing direction**: forward.
VII. Interval: 20.
VIII. Resource sharing fraction**: 0.2.
IX. Generations**: 500.
X. Stall limit: 50.
XI. Creation function: treesocialcreationuniform.

Inspired by the social relations and resource-sharing behaviors observed in tree communities, the TSR algorithm iteratively refined the network's parameters. This ensured optimal concentrations of medium components to maximize antibiotic production. By employing TSR, the optimization process mimicked natural selection and accounted for complex, non-linear interactions within the network, resulting in a robust model for predicting and enhancing Actinomycin V yield [7], [9].

5|Results and Discussion

The relationship between medium component concentrations and antibiotic yield was examined using RSM, which indicated a poor fit with an overall regression coefficient (R^2) of 0.76. Significant effects were identified for four model coefficients: the linear effect of glucose, the quadratic effect of soybean meal, and the interaction effects between MgSO4–glucose and NaCl–glucose (*Table 2*). The antibiotic yield predicted by RSM (*Eq. (2)*) differed significantly from the experimentally observed values (*Table 1*).

To improve the optimization of *Eq. (2)*, we employed the TSR optimization algorithm instead of genetic algorithms. Despite this enhancement, the maximum predicted antibiotic yield at the optimized conditions (best individual not shown) was 326.5 mg/L. Experimental validation, conducted in duplicate, with the optimized concentrations in the basal medium, yielded an average recovery of 324 mg/L of actinomycin V, closely matching the predicted value.

Eq. (3) represents the MATLAB implementation of the trained feed-forward ANN model (graphical representation in *Fig. 2*), which correlates medium component concentrations with antibiotic yield. In this model, `act` denotes the network, `act.IW` represents the input layer weights, `act.LW{2,1}` corresponds to the hidden layer weights, and `act.LW{3,2}` represents the output layer weights. The transfer functions used are `pur` and `tanS`. The network's layer weights were determined after training and validation. A comparison of antibiotic yield predictions from RSM and ANN with experimentally observed values (*Fig. 3*) demonstrates that the ANN network's predictions are significantly closer to the observed yields compared to those from RSM.

$$y = \sum_{i=1}^{q} \beta_i x_i + \sum\sum \beta_{ij} x_i x_j + \sum \sum_{i<j<k}^{q} \beta_{ijk} x_i x_j x_k. \quad \textbf{(2)}$$

$$\mathrm{Net} = \mathrm{pur}(\mathrm{act.LW\{3,2\}}* \tanS(\mathrm{act.LW\{2,1\}}* \tanS(\mathrm{act.LW\{1,1\}}*p+\mathrm{act.b\{1\}})+\mathrm{act.b\{2\}})+\mathrm{act.b\{3\}}). \quad \textbf{(3)}$$

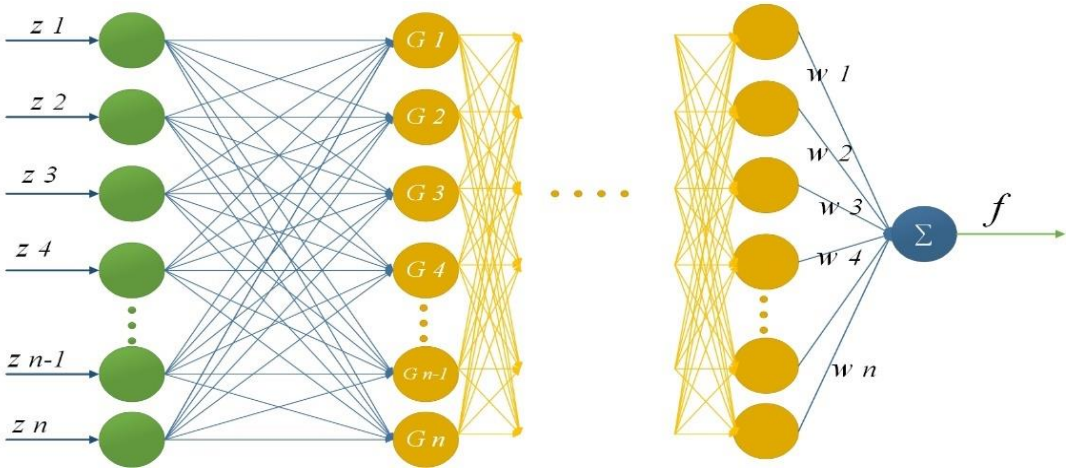

Fig. 2. ANN style.

5.1 | Optimization of the Neural Network Using TSR Optimization Algorithm

The algebraic form of *Eq. (3)* was utilized as the fitness function for optimization with the TSR optimization algorithm. Employing a population of 270, the neural network responses efficiently converged to optimal values within just eleven generations (*Fig. 4*). The TSR-optimized concentrations of medium components resulted in a maximum antibiotic yield of 452 mg/L, with the optimal conditions being MgSO4 at 3.657 g/L, NaCl at 1.9012 g/L, glucose at 8.836 g/L, soybean meal at 20.1976 g/L, and CaCO3 at 13.0842 g/L [15].

In contrast, the quadratic polynomial model (*Eq. (2)*), optimized using TSR, predicted a lower yield of 326.5 mg/L at optimal concentrations. This highlights the superiority of neural networks over polynomial models for predicting antibiotic yield based on medium component concentrations. The TSR-optimized solution, confirmed experimentally, yielded 443 (±5) mg/L of actinomycin V, closely aligning with the TSR-predicted yield of 452 mg/L. This optimization led to a nearly fourfold increase in actinomycin V yield (110 mg/L to 443 mg/L) compared to the unoptimized medium. When comparing the outputs from RSM and ANN, the ANN provided a 39.4% improvement in performance [2].

Fig. 5 illustrates how antibiotic yield varies with changes in individual medium component concentrations while keeping other components at their central values. The data show that lower concentrations of glucose and CaCO3, moderate levels of soybean meal, and higher concentrations of NaCl and MgSO4 initially increased antibiotic production. However, exceeding optimal levels of these components resulted in a decrease in antibiotic yield [9].

6 | Discussion

The newly isolated strain 'S. triostinicus' was initially found to produce actinomycin V at a concentration of 110 mg/L in an unoptimized production medium. Using RSM, production levels were increased to 325 mg/L. Further enhancement was achieved by applying a combination of ANN and the TSR optimization algorithm, which boosted the actinomycin V yield to 443 mg/L.

An optimized medium composition for actinomycin V production using 'S. triostinicus' was developed through artificial intelligence-based modeling and compared to the RSM optimization method. The ANN model demonstrated excellent prediction accuracy and generalization capabilities. A comparison between RSM and ANN results confirmed the efficacy of the neural network as an empirical modeling tool. The ANN fit the training data exceptionally well and provided accurate predictions for validation data.

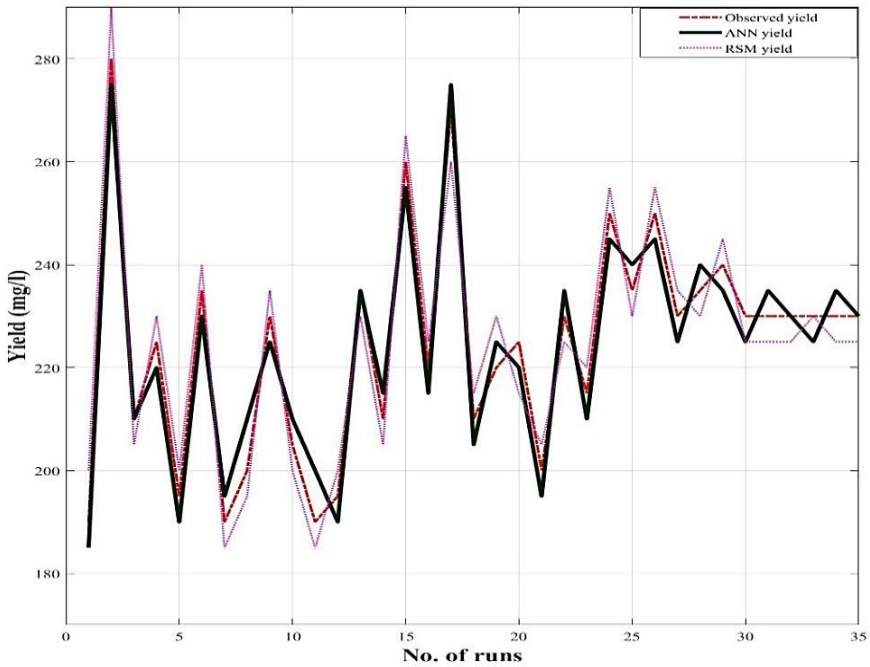

Fig. 3. ANN and RSM experimental results.

The ANN model was further validated by assessing how variations in the concentration of individual medium components affected antibiotic yield. The model's outputs closely matched the experimental results, reflecting the role of medium components in actinomycin V production. The biosynthesis of actinomycin V involves the tryptophan pathway, which is influenced by various amino acids such as valine, histidine, ornithine, tryptophan, and leucine [16]. As a complex nitrogen source, soybean meal contains multiple amino acids, including lysine, methionine, threonine, tryptophan, aspartic acid, glutamine, proline, alanine, valine, and isoleucine. Tryptophan in soybean meal enhances antibiotic production up to a certain concentration level.

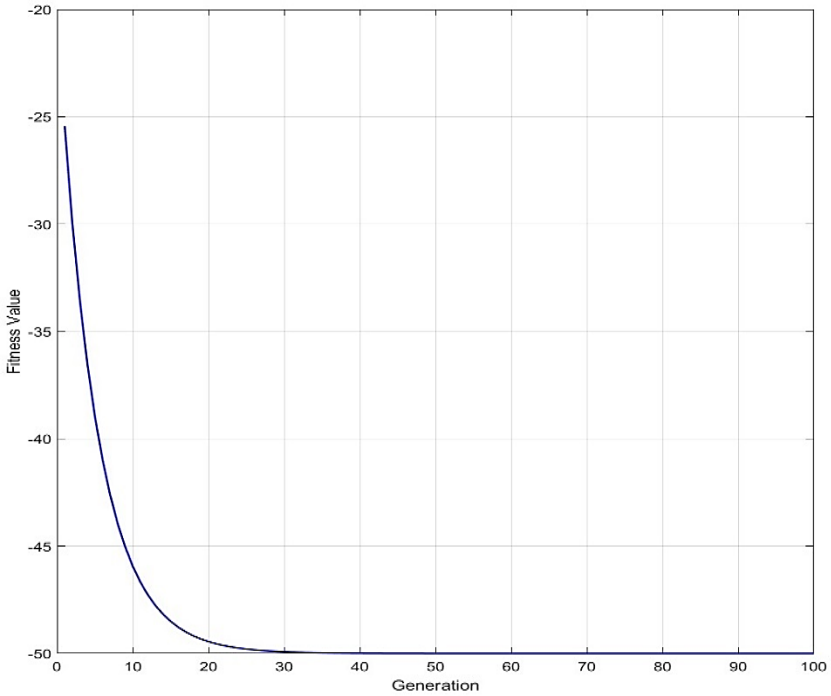

Fig. 4. Illustrates the gradual improvement in the performance of TSR generations until the optimal solution is achieved.

However, the presence of other amino acids can lead to a decrease in actinomycin V production beyond an optimal level. For example, it was reported by Foster and Katz [17] that more than 50% repression of the tryptophan oxygenase enzyme was observed due to the presence of glutamate, aspartate, methionine, proline, and valine. This enzyme is crucial in converting tryptophan to N-formyl kynurenine in the tryptophan metabolism pathway.

Additionally, high glucose concentrations have been shown to repress the formation of phenoxazinone synthetase, an enzyme necessary for actinomycin synthesis [18]. Various cations such as Mg^{2+}, Fe^{2+}, Zn^{2+}, Cu^{2+}, Mn^{2+}, and Co^{2+} are known to stimulate antibiotic production [16], and a similar stimulatory effect of Mg^{2+} was observed in this study.

This study demonstrates that the ANN-TSR hybrid approach significantly enhances actinomycin V production. The methodology presented is broadly applicable and can be extended to model and optimize other bioprocesses effectively.

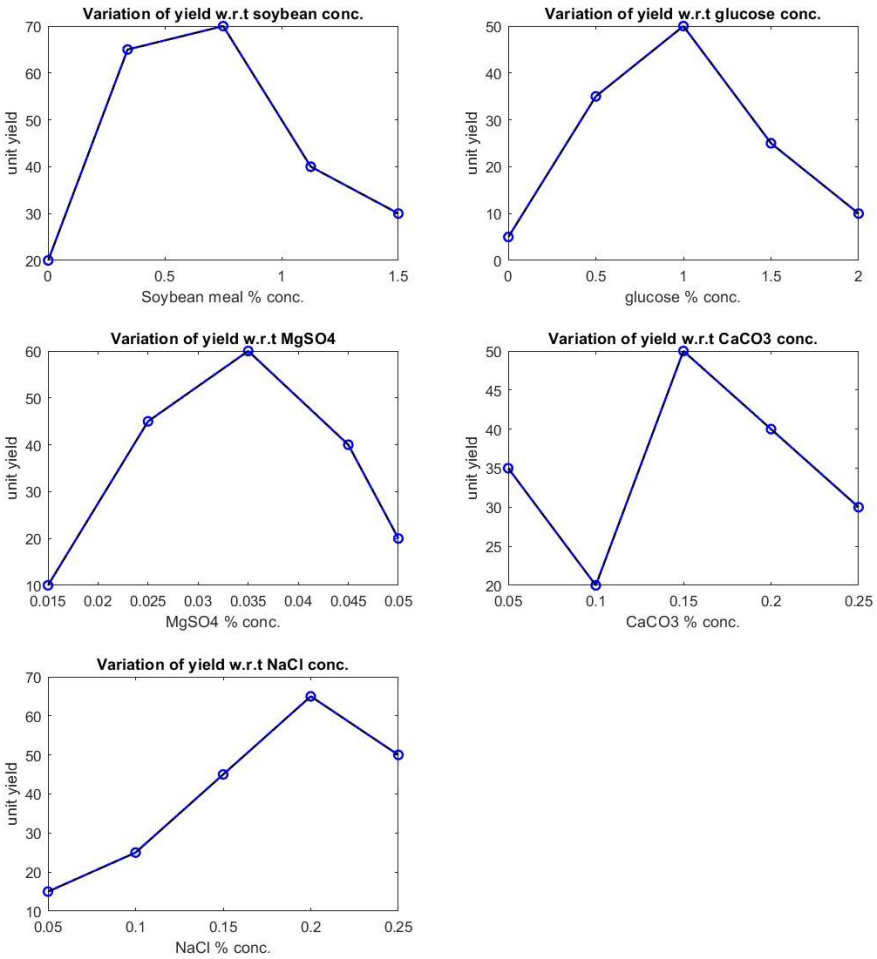

Fig. 5. Graphs of various medium components, with all other components maintained at zero level.

7 | Conclusion

The hybrid optimization approach combining ANNs and the TSR algorithm has demonstrated significant improvements in Actinomycin V production by Streptomyces triostinicus. This innovative methodology not only outperforms traditional techniques like RSM but also offers a scalable framework for optimizing complex industrial bioprocesses. The ANN-TSR model's ability to accurately predict and enhance yields from 110 mg/L to 443 mg/L underscores its potential as a powerful tool in bioprocess engineering.

The study confirms that integrating advanced machine learning with nature-inspired algorithms can effectively address the challenges of nonlinear interactions in bioprocess optimization. By systematically analyzing medium components and employing a robust ANN-TSR framework, this research opens new avenues for optimizing various industrial fermentation processes, offering substantial benefits in both yield and cost efficiency. The findings serve as a foundation for further application of these advanced methods across different biotechnological domains.

Author Contributions

All authors contributed to the study's conception and design. Marzieh Lamtar-Gholipoor performed material preparation, data collection, and Software: conceptualization, Visualization, Writing - review & editing by Soheil Fakhri. Mahmoud Alimoradi wrote the first draft of the manuscript, Writing the original draft, Supervision Methodology, and software. All authors read and approved the final manuscript.

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Data Availability

The data supporting the findings of this study are available upon request. Interested researchers can obtain the data by contacting the authors via email.

Conflict of Interest

The authors stated that there are no conflicts of interest regarding the publication of this article.

References

[1] Giera, M., Aisporna, A., Uritboonthai, W., & Siuzdak, G. (2024). The hidden impact of in-source fragmentation in metabolic and chemical mass spectrometry data interpretation. *Nature metabolism*, 1–2. https://doi.org/10.1038/s42255-024-01076-x

[2] Li, Y., Cavet, G., Zare, R. N., & Driver, T. (2024). Fragment correlation mass spectrometry: Determining the structures of biopolymers in a complex mixture without isolating individual components. *Proceedings of the national academy of sciences*, *121*(32), e2409676121. https://doi.org/10.1073/pnas.2409676121

[3] Azgomi, H., Haredasht, F. R., & Motlagh, M. R. S. (2023). Diagnosis of some apple fruit diseases by using image processing and artificial neural network. *Food control*, *145*, 109484. https://doi.org/10.1016/j.foodcont.2022.109484

[4] Daliri, A., Asghari, A., Azgomi, H., & Alimoradi, M. (2022). The water optimization algorithm: a novel metaheuristic for solving optimization problems. *Applied intelligence*, *52*(15), 17990–18029. DOI:10.1007/s10489-022-03397-4.

[5] Alimoradi, M., Azgomi, H., & Asghari, A. (2022). Trees social relations optimization algorithm: a new swarm-based metaheuristic technique to solve continuous and discrete optimization problems. *Mathematics and computers in simulation*, *194*, 629–664. https://doi.org/10.1016/j.matcom.2021.12.010

[6] Fakheri, S., Alimoradi, M., & Yamaghani, M. R. (2024). *Colour image multilevel thresholding segmentation using trees social relationship algorithm*. https://doi.org/10.21203/rs.3.rs-4479475/v1%0A%0A

[7] Daliri, A., Alimoradi, M., Zabihimayvan, M., & Sadeghi, R. (2024). World Hyper-Heuristic: A novel reinforcement learning approach for dynamic exploration and exploitation. *Expert systems with applications*, *244*, 122931. https://doi.org/10.1016/j.eswa.2023.122931

[8] Alimoradi, M., Zabihimayvan, M., Daliri, A., Sledzik, R., & Sadeghi, R. (2022). Deep neural classification of darknet traffic. In *Artificial intelligence research and development* (pp. 105–114). IOS Press. DOI: 10.3233/FAIA220323

[9] Alimoradi, M., Fakheri, S., & others. (2024). A novel metaheuristic approach inspired by trees social relationships and models for fermentation medium. *Metaheuristic algorithms with applications*, *1*(1), 1–11. https://orcid.org/0000-0001-5751-6997

[10] Barbosa, M. J., Janssen, M., Südfeld, C., D'Adamo, S., & Wijffels, R. H. (2023). Hypes, hopes, and the way forward for microalgal biotechnology. *Trends in biotechnology*, *41*(3), 452–471. https://doi.org/10.1016/j.tibtech.2022.12.017

[11] Graver, B. A., Chakravarty, N., & Solomon, K. V. (2024). Prokaryotic Argonautes for in vivo biotechnology and molecular diagnostics. *Trends in biotechnology*, *42*(1), 61–73. https://doi.org/10.1016/j.tibtech.2023.06.010

[12] Liao, C., Xiao, S., & Wang, X. (2023). Bench-to-bedside: Translational development landscape of biotechnology in healthcare. *Health sciences review*, *7*, 100097. https://doi.org/10.1016/j.hsr.2023.100097

[13] Alimoradi, M., Daliri, A., Zabihimayvan, M., & Sadeghi, R. (2024). *Statistic deviation mode balancer (SDMB): A novel sampling algorithm for imbalanced data*. https://doi.org/10.21203/rs.3.rs-4009264/v1%0A%0A

[14] Holzinger, A., Keiblinger, K., Holub, P., Zatloukal, K., & Müller, H. (2023). AI for life: Trends in artificial intelligence for biotechnology. *New biotechnology*, *74*, 16–24. https://doi.org/10.1016/j.nbt.2023.02.001

[15] Kamiki, R., Kubo, T., & Satoh, K. (2023). Addition--fragmentation ring-opening polymerization of bio-based thiocarbonyl l-lactide for dual degradable vinyl copolymers. *Macromolecular rapid communications*, *44*(2), 2200537. https://doi.org/10.1002/marc.202200537

[16] Williams, W. K., & Katz, E. (1977). Development of a chemically defined medium for the synthesis of actinomycin D by Streptomyces parvulus. *Antimicrobial agents and chemotherapy*, *11*(2), 281–290. https://doi.org/10.1128/aac.11.2.281

[17] Foster, J. W., & Katz, E. (1981). Control of actinomycin D biosynthesis in Streptomyces parvullus: regulation of tryptophan oxygenase activity. *Journal of bacteriology*, *148*(2), 670–677. https://doi.org/10.1128/jb.148.2.670-677.1981

[18] Gallo, M., & Katz, E. (1972). Regulation of secondary metabolite biosynthesis: catabolite repression of phenoxazinone synthase and actinomycin formation by glucose. *Journal of bacteriology*, *109*(2), 659–667. https://doi.org/10.1128/jb.109.2.659-667.1972